XXI iwCLL 2025
Poster No. 1001

# IgM Monoclonal Gammopathy Arises Directly from MBL/CLL Cells: Evidence from Proteomic Analyses

Uday Aditya Sarkar[1,2,3], Dylan Tabang[4,5], Colin McNulty[1], Sabine Allam[1,3], Ayazhan Umutbayeva[1], Stacey Fernandes[1,3], Mariia Mikhaleva[1], Roberta Santos Azevedo[1], Zainab Wurie[4], Ava Bidgoli[1], Natalie O. Onofri[1], Joseph Flinn[1], Kiyomi Mashima[1,2,3], Ingrid Lourme[4], Elisavet Vlachonikola[7], Binu K. Sasi[1,2,3], Paulina Predko[1], Andreas Agathangelidis[6], Kenneth Parker[4], Kostas Stamatopoulos[7], Irene Ghobrial[1,2,3], Jennifer R. Brown[1,2,3], Hanno Steen[4,5], Paolo Ghia[1,2,3,8,9]

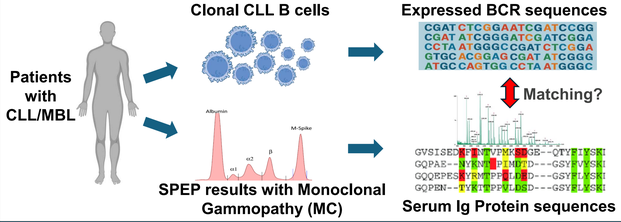
1 Department of Medical Oncology, Dana-Farber Cancer Institute, Boston, MA, USA 2 Department of Medicine, Harvard Medical School, Boston, MA, USA 3 Cancer Program, Broad Institute of MIT & Harvard, Cambridge, MA, USA 4 Department of Pathology, Boston Children's Hospital, Boston, MA, USA 5 Department of Pathology, Harvard Medical School, Boston, MA, USA 6 Division of Genetics and Biotechnology, Department of Biology, National and Kapodistrian University, Athens, Greece 7 Institute of Applied Biosciences, Centre for Research and Technology Hellas, Thessaloniki, Greece 8 Università Vita-Salute San Rffaele, Milano, Italy 9 IRCCS Ospedale San Raffaele, Milano, Italy

## Background & Aim

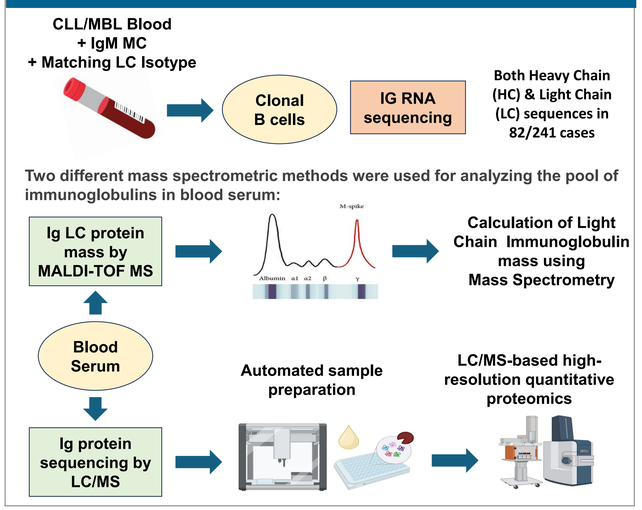
Chronic Lymphocytic Leukemia (CLL) is the most frequent leukemia in adults and is typically preceded by a pre-malignant condition called Monoclonal B cell Lymphocytosis (MBL), which is detected in up to 10% of otherwise healthy individuals. Monoclonal gammopathy of undetermined significance (MGUS) of the IgM isotype is also a common pre-neoplastic condition concerning healthy individuals, also requiring regular clinical follow-up. MGUS can be detected before, after, or simultaneously with an MBL/CLL diagnosis.

Using innovative Ig proteomics methods in our study, we aimed to explore whether the clonal CLL B cells could be responsible for producing the serum IgM monoclonal component (MC), thus being directly responsible for the development of IgM MGUS, or if the latter should be considered a manifestation of another concomitant disease, given the increased risk of other neoplasias in patients with MBL/CLL.

## Rationale




## Methods




## Results

**Table 1: CLL/MBL patients characteristics**

| Characteristics | Number (%) |
|---|---|
| Total Patients (N) | 82 |
| Male | 53 (64.6) |
| Female | 29 (35.4) |
| Age (years) | |
| Mean ± SD | 66.61 ± 12.19 |
| Median; Range | 68; 27-91 |
| Diagnosis (at sample time) | |
| CLL | 79 (96.3) |
| MBL | 3 (3.7) |
| M-Spike (SPEP Results) | |
| IgM Kappa | 56 (68.3) |
| IgM Lambda | 21 (25.6) |
| Free IgM | 3 (3.7) |
| Free Lambda | 2 (2.4) |

**Figure 1: Observed MW from MALDI-TOF MS correlates with calculated MW from expressed LC sequence from clonal cells**

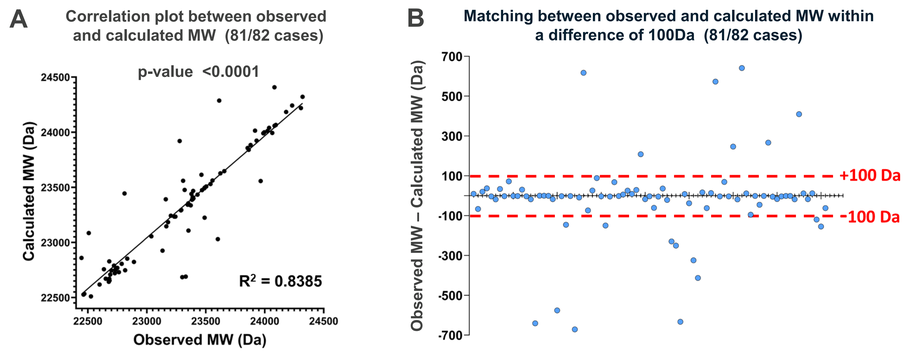



**Figure 2A: LC/MS based quantitative proteomics was used to sequence the immunoglobulins present in the blood serum**

| ID Code | IGHV Gene | IGKV Gene | IGLV Gene | Note |
|---|---|---|---|---|
| PG-0020 | IGHV3-72*01 F | IGKV4-1*01 F | | Both IGHV and IGKV matched |
| PG-0021 | IGHV4-30-2*07 | IGKV1-33*01 F | | Only IGKV matched |
| PG-0022 | IGHV3-13*05 F | | IGLV1-51*01 F | Only IGLV matched |
| PG-0023 | IGHV3-21*01 F | | IGLV3-21*04 F | No match |

IG sequences expressed by CLL/MBL cells at RNA level

LC/MS allowed to sequence the most abundant Ig proteins.

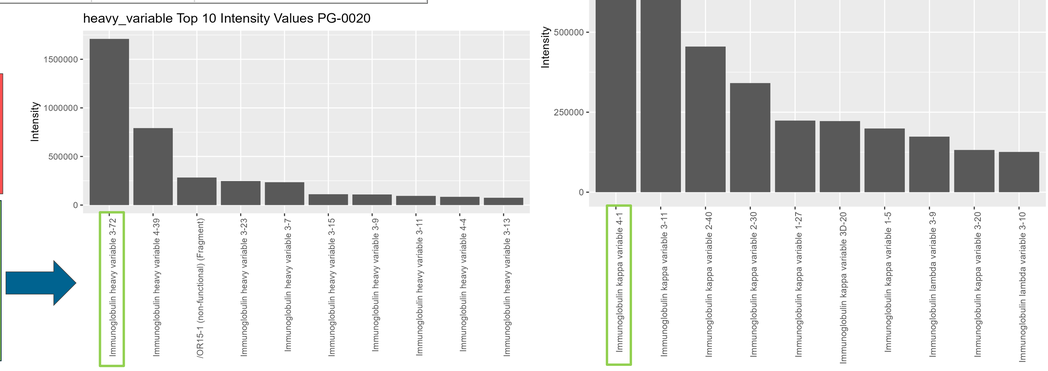
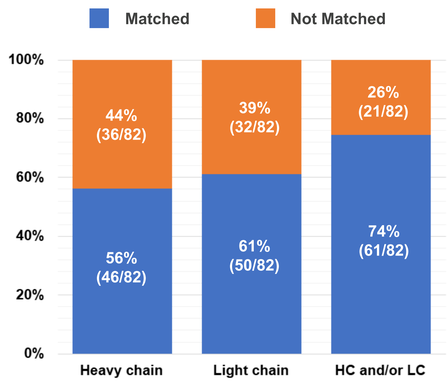



**Figure 2B: 74% (61/82) of the IG Heavy and/or Light variable genes match the Ig proteins found in the sera of patients with CLL/MBL**




## Conclusions

- The serum Ig LC mass measured by MALDI-TOF MS matched the calculated mass from the expressed clonal Ig in 76% (62/82) of the MBL/CLL cases.
- In 74% (61/82) of the cases, quantitative LC/MS proteomics confirmed the presence of matched IGHV (56%) and IGKV/IGLV (61%) genes in the sera of patients with CLL/MBL.
- **In the vast majority of cases of CLL/MBL, clonal B cells produce the serum IgM monoclonal component. The lack in matching is most likely due to the low levels of MC among the serum Immunoglobulins.**

## Future Directions

- Work in future aims to expand the cohort to include more patients with MBL to confirm our findings in the pre-leukemic condition as well.
- We will also include CLL/MBL cases with IgG and IgA monoclonal component in the serum.
- The matching between the RNA and the protein sequences will be refined by attempting to identify the peptides corresponding to the unique CDR3 portion of the Ig expressed by clonal cells in each patient.

## Funding

The work for this project has been primarily supported by the funding received from the Center for Early Detection and Interception of Blood Cancers (CEDI-BC) grant 2023-24 awarded to Paolo Ghia and Irene Ghobrial.

## Contact Information

Uday Aditya Sarkar, Ph.D.
uday_sarkar@dfci.harvard.edu

Corresponding Author:
Paolo Ghia, M.D., Ph.D.
paolop_ghia@dfci.harvard.edu

CLL Center, Department of Medical Oncology, Dana-Farber Cancer Institute, 450 Brookline Avenue, Boston, MA 02215